
KARYOTYPE VARIATION IN *PANCRATIUM HIRTUM* A. CHEV. (AMARYLLIDACEAE)¹

S. O. Oyewole²

ABSTRACT

*Natural populations of *Pancratium hirtum* A. Chev. from different ecological niches show definable morphological variation. Samples were grown in an experimental garden and investigated karyotypically. Five morphological variants (morphotypes) were identified and were confirmed as showing karyotype differences. Karyotype differentiation involves at least: (1) chromosomal breakages, and (2) differences in the total length of chromatin material per nucleus. The chromosome basic number of $x = 11$ is confirmed for the species, while the presence of accessory chromosomes in one morphotype is reported for the first time in the genus. Population divergence is less pronounced than karyotypic divergence.*

Pancratium L. is represented in West Tropical Africa by two species, *P. hirtum* A. Chev. and *P. trianthum* Herb. Their taxonomy was well documented by Morton (1965), who reported the somatic chromosome number of $2n = 22$ for them. Morton did not include analysis of the karyotypes. *Pancratium hirtum*, with large chromosomes, is especially suitable for study of chromosome morphology.

MATERIALS AND METHODS

Pancratium hirtum grows in a variety of niches in savanna vegetation, where it exhibits minor but definable differences in leaf size, leaf color, extent of pubescence, length and form of the peduncle, and texture of the outermost tunic of the bulb (Table 1).

Five morphological groups (morphotypes, Fig. 1) were recognized during field study and samples were collected. Not fewer than 40 bulbs of each morphotype were grown separately on adjacent beds in the experimental garden. Their habitats are described in Table 2.

Each bulb in each morphotype was ex-

amined cytologically using root tip squashes as outlined in Darlington & LaCour (1969). Chromosome counts were made from several metaphase plates in each preparation. Measurement of chromosomes using calibrated micrometer eyepiece graticule was impracticable due to their unusual lengths. Photographs of metaphase plates were taken at $\times 7.5$ ocular and $\times 40$ objective of the Olympus (Vanox model) Research Microscope. Chromosomes were measured from the photomicrographs. The measurements were pooled from 15–20 complements for each morphotype, and average lengths were determined. Chromosome morphological determinations were according to Levan et al. (1964) as modified by Adhikary (1974). Evidence of chromosomal changes was manifested in unequalness of members of homologous chromosome pairs (such changes usually affect one arm of a member). In such instances, the unaffected arm length was employed to identify the members, and the chromosome index was based solely on the length of the longer member of the pair. Idiograms were constructed from enlarged photomicrographs.

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² Department of Biological Sciences, University of Ilorin, P.M.B. 1515, Ilorin, Nigeria.

RESULTS

Karyotype data are summarized in Table 3. Figure 2 shows metaphase plates of the somatic complements, and Figure 3 presents the idiograms. All morphotypes have a somatic chromosome number of $2n = 22$, except E, in which the complement is $2n = 22 + 4$ B-chromosomes.

Morphotype A. The chromosomes vary in length from $5.5\text{ }\mu\text{m}$ to $19\text{ }\mu\text{m}$, with a total length of chromatin material of $216.5 \pm 6.5\text{ }\mu\text{m}$. The complement (Figs. 4A, 5A) consists of four pairs with median to submedian centromeres (1st, 2nd, 3rd, and 11th), one telocentric pair (8th), and six pairs with terminal to subterminal centromeres (4th–7th, 9th and 10th). The longest two pairs have unequal members, one member of each having lost a portion of its long arm.

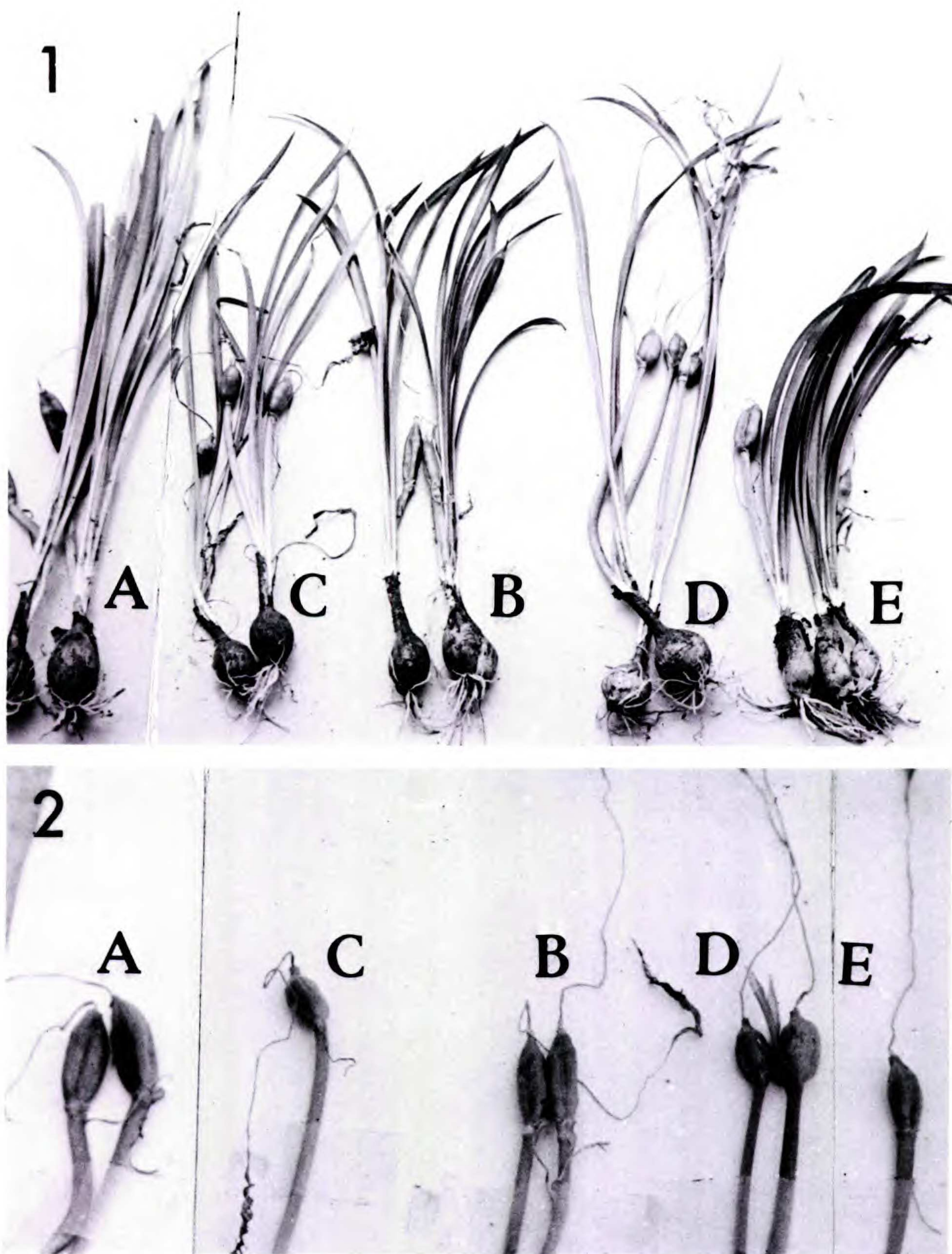
Morphotype B. Chromosomes vary in length from $11.38\text{ }\mu\text{m}$ to $41.75\text{ }\mu\text{m}$, with an average total chromatin length of $439.5 \pm 18.5\text{ }\mu\text{m}$. The complement (Figs. 4B, 5B) consists of three pairs (1st, 2nd, and 11th) with median to submedian centromeres, and eight (3rd–10th) with terminal to subterminal centromeres. The second- and third-longest pairs have unequal members: a member of the former having lost a portion of the long arm, while one member of the latter lost a portion of its short arm.

Morphotype C. Chromosomes vary in length from $6.3\text{ }\mu\text{m}$ to $19.14\text{ }\mu\text{m}$, and the total chromatin length averages $237.3 \pm 23.3\text{ }\mu\text{m}$. The complement (Figs. 4C, 5C) consists of three pairs (1st, 2nd, and 11th) with median to submedian centromeres and eight pairs (3rd–10th) with terminal to subterminal centromeres. The third-longest pair has unequal members—the shorter member has a shorter second arm. One member of the shortest pair also shows loss of a portion of one arm.

Morphotype D. Chromosome length varies between $8.6\text{ }\mu\text{m}$ and $27.3\text{ }\mu\text{m}$, with an average total chromatin length of $340.4 \pm 49.8\text{ }\mu\text{m}$. The complement (Figs. 4D, 5D)

TABLE 1. Summary of the external morphology of *Pancratium hirtum* morphotypes.

	A	B	C	D	E
Peduncle	4–7 cm long, flattened, puberulous	2–4.5 cm long, elliptic, fluted, faintly puberulous	4–7 cm long, cylindrical, glabrous	10–13 cm long, cylindrical, cal, glabrous	4–6 cm long, slightly flattened, shortly puberulous
Fruit	2.5–3.5 cm long, 3–3.6 cm circumference, oblong-elliptic, obtuse apex, trigonal in transverse section, green, glabrous, receptacle 1–3 mm high	2–4 cm long, 2–3.3 cm circumference, lanceolate, obtuse apex, trigonal in transverse section, green, glabrous, receptacle 1–3 mm high	1.7–2 cm long, 2–3.6 cm circumference, oblong, apex 4–5 mm high, trigonal in transverse section, green, glabrous, receptacle 1–3 mm high	1.4–2 cm long, 2.5–3.5 cm circumference, cylindrical, apex 2–4 mm high, trigonal in transverse section, green, glabrous, receptacle 1–3 mm high	2–2.5 cm long, 3–4 cm circumference, spindle shaped, apex 1–5 mm high, obtusely trigonal, green, glabrous, receptacle 1–3 mm high
Leaf	light green, 26–33 cm long, 9–15 mm wide	dark green, 21–30 cm long, 4–9 mm wide	light green, 22–31 cm long, 5–9 mm wide	light green, 24–35 cm long, 5–9 mm wide	dark green, 20–26 cm long, 7–14 mm wide
Bract	single, deciduous, about half-encircling, single lipped, yellowish green	single, deciduous, half-encircling, single lipped, green	single, deciduous, half-encircling, single lipped, light green	single, persistent until fruit ripening, fully encircling, 2-lipped, green	single, deciduous, half-encircling, single-lipped, dark green



FIGURES 1, 2.—1. *Pancratium hirtum* morphotypes with mature fruits, $\times 0.2$.—2. Mature fruits, with peduncles, of *Pancratium hirtum* morphotypes, $\times 0.4$.

consists of five pairs (1st–3rd, 10th, and 11th) with median to submedian centromeres and six pairs (4th–9th) with terminal centromeres. Five of the eleven pairs (1st, 2nd, 3rd, 8th, and 10th) show evidence of loss of portions from one member of each pair. First and 10th pairs show loss in the long arm; the second and third pairs show loss in the short arm,

and the eighth shows loss in one of the two equal arms.

Morphotype E. The autosomes vary in length from $5.6\ \mu\text{m}$ to $17.9\ \mu\text{m}$, and the B-chromosomes vary between $1\ \mu\text{m}$ and $1.5\ \mu\text{m}$. The average total chromatin length is $219.14 \pm 2.5\ \mu\text{m}$. The complement (Figs

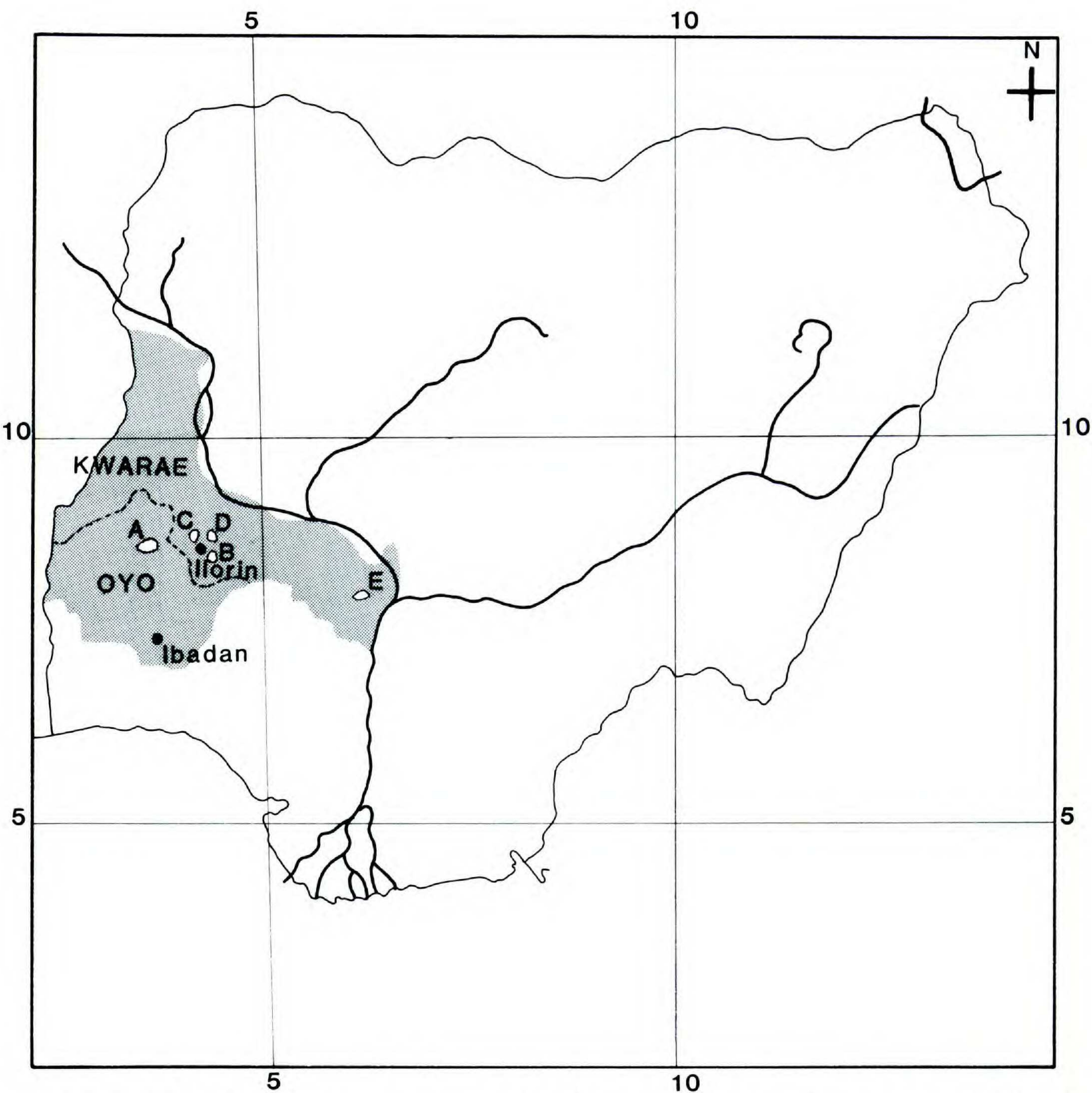


FIGURE 3. Map of Nigeria showing locations where *Pancratium hirtum* populations were sampled in Oyo and Kwara States. Broken line = state boundary; circles = state capital city; solid lines = rivers.

4E, 5E) consists of five pairs (1st, 2nd, 4th, 8th, and 11th) with median to submedian centromeres, and six pairs (3rd, 5th, 6th, 7th, 9th, and 10th) with terminal to subterminal centromeres. All the B-chromosomes are telocentric. One member of the second-longest autosomal pair has chromosome loss in the long arm, while one member of each of the eighth and ninth pairs shows loss in the short arm.

DISCUSSION

Previous chromosome counts in *Pancratium* show a chromosome number of $2n =$

22 (Ponnamma, 1978; Lakshmi, 1980). This number is confirmed here except that the presence of accessory chromosomes had not been reported previously in the genus.

The five karyotypes share a basic plan: the largest two and the smallest pairs of chromosomes are metacentric while all the others are acrocentric. Deviations from this plan consist of increase in the number of metacentrics (A, D, and E with an increase of one, two, and two pairs, respectively). Apart from these, there is evidence of structural changes in the chromosomes as a result of loss or gain of segments. This is common to all the karyo-

TABLE 2. Sources of material of *Pancratium hirtum*.

Mor- pho- types	Collection Site	Herbarium Voucher	Habitat
A	Iseyin-Igbetti about 150 km northwest of Ibadan	S00/2106 in University of Ilorin Herbarium (IUH)	deciduous woodland, in dark humus, under the shade of trees such as <i>Butyrospermum paradoxum</i> , <i>Lophira lanceolata</i> , etc.
B	Affon, 25 km southeast of Ilorin	S00/2199 in IUH	disturbed woodland, in dark humus soil on shallow inselbergs and foot of rocky hill under the shade of <i>Parkia biglobosa</i>
C	Shao, 22 km northwest of Ilorin	S00/2200 in IUH	open and exposed brown soil of old mats of <i>Afrotrilepis pilosa</i> on rock outcrops in savanna grassland
D	Oke-Oyi, about 20 km north of Ilorin	S00/2201 in IUH	open brown humus soil among rock boulders in savanna grassland
E	Okene-Lokoja Road, about 50 km from Okene	S00/2202 in IUH	dark brown gravelly clay-loam in floodplains in savanna woodland under stands of <i>Khaya senegalensis</i>

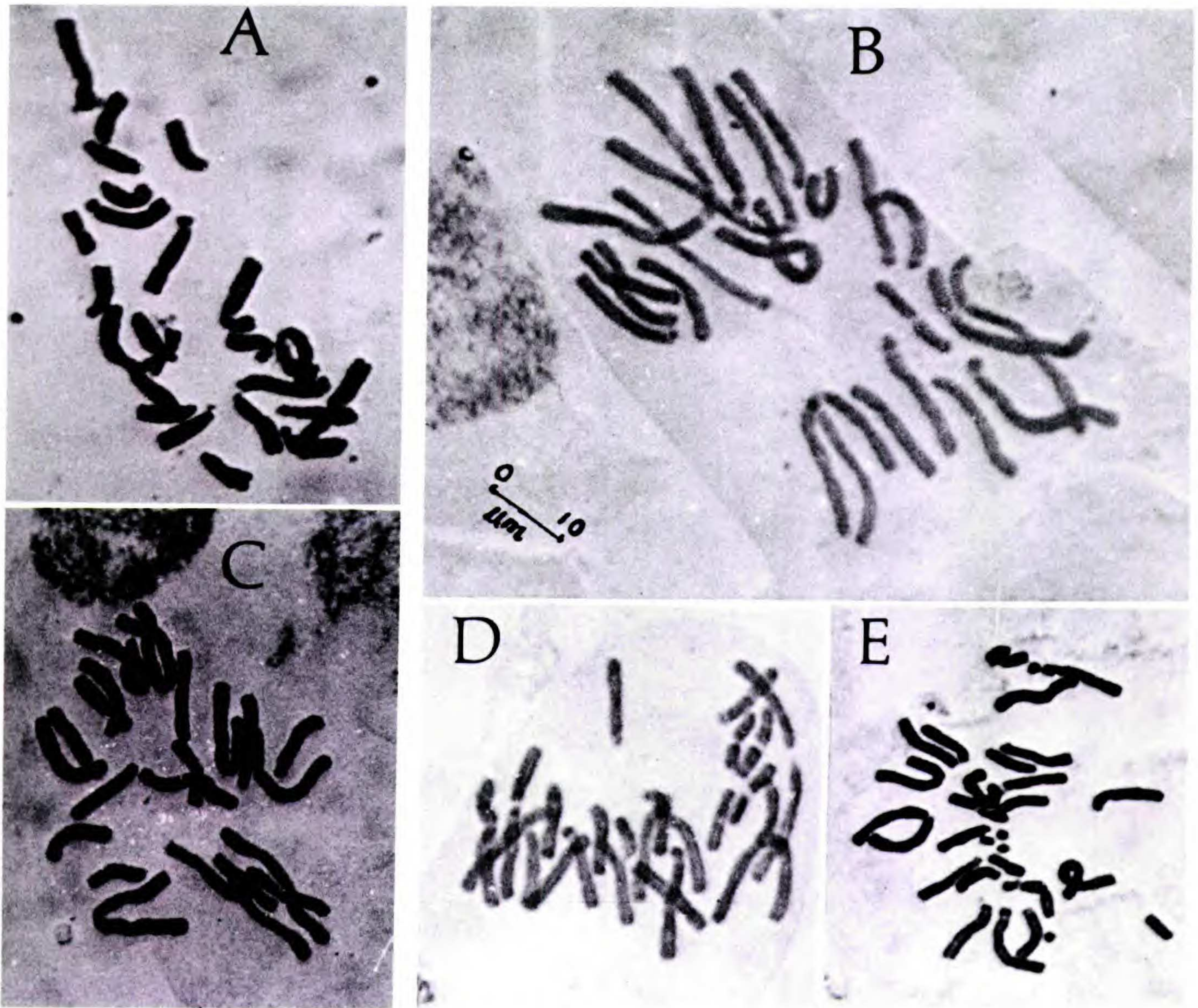


FIGURE 4. Somatic metaphase complements of *Pancratium hirtum* morphotypes.

TABLE 3. Karyotype data for *Pancratium hirtum* (chromosome length in μm).

Homologues	A	B	C	D	E
1	chromosome length	19.0	19.14	27.25	17.88
	<i>r</i> -value	1.1	1.0	1.17	1.37
	centromere location	median	median	median	median
2	chromosome length	16.0	15.42	25.80	14.5
	<i>r</i> -value	1.0	1.76	1.48	1.7
	centromere location	median	median	median	median
3	chromosome length	12.25	13.92	21.06	12.63
	<i>r</i> -value	2.3	3.64	2.56	11.25
	centromere location	submedian	subterminal	submedian	terminal
4	chromosome length	10.75	11.92	17.75	11.88
	<i>r</i> -value	9.75	10.92	10.83	1.97
	centromere location	terminal	terminal	terminal	submedian
5	chromosome length	9.0	9.75	15.0	9.0
	<i>r</i> -value	8.0	13.63	10.25	35.0
	centromere location	terminal	terminal	terminal	terminal
6	chromosome length	7.5	9.25	13.0	8.25
	<i>r</i> -value	14.0	21.2	11.0	15.5
	centromere location	terminal	terminal	terminal	terminal
7	chromosome length	7.5	9.0	12.0	8.25
	<i>r</i> -value	14.0	8.0	13.4	7.25
	centromere location	terminal	terminal	terminal	terminal
8	chromosome length	7.0	8.58	10.92	7.38
	<i>r</i> -value	0	7.58	17.71	2.04
	centromere location	terminal	terminal	terminal	submedian
9	chromosome length	7.0	8.0	10.75	6.63
	<i>r</i> -value	6.0	11.0	13.33	3.0
	centromere location	subterminal	terminal	terminal	subterminal
10	chromosome length	6.75	7.33	10.0	6.25
	<i>r</i> -value	26.0	21.0	1.07	4.0
	centromere location	terminal	terminal	median	subterminal
11	chromosome length	5.5	6.33	8.67	5.67
	<i>r</i> -value	1.2	1.0	1.0	1.2
	centromere location	median	median	median	median
Accessory chromosomes		—	—	—	1.5, 1.0
		—	—	—	1.0, 1.0
	total chromatin length	216.5 \pm 6.5	237.3 \pm 23.3	340.4 \pm 49.8	219.14 \pm 2.5

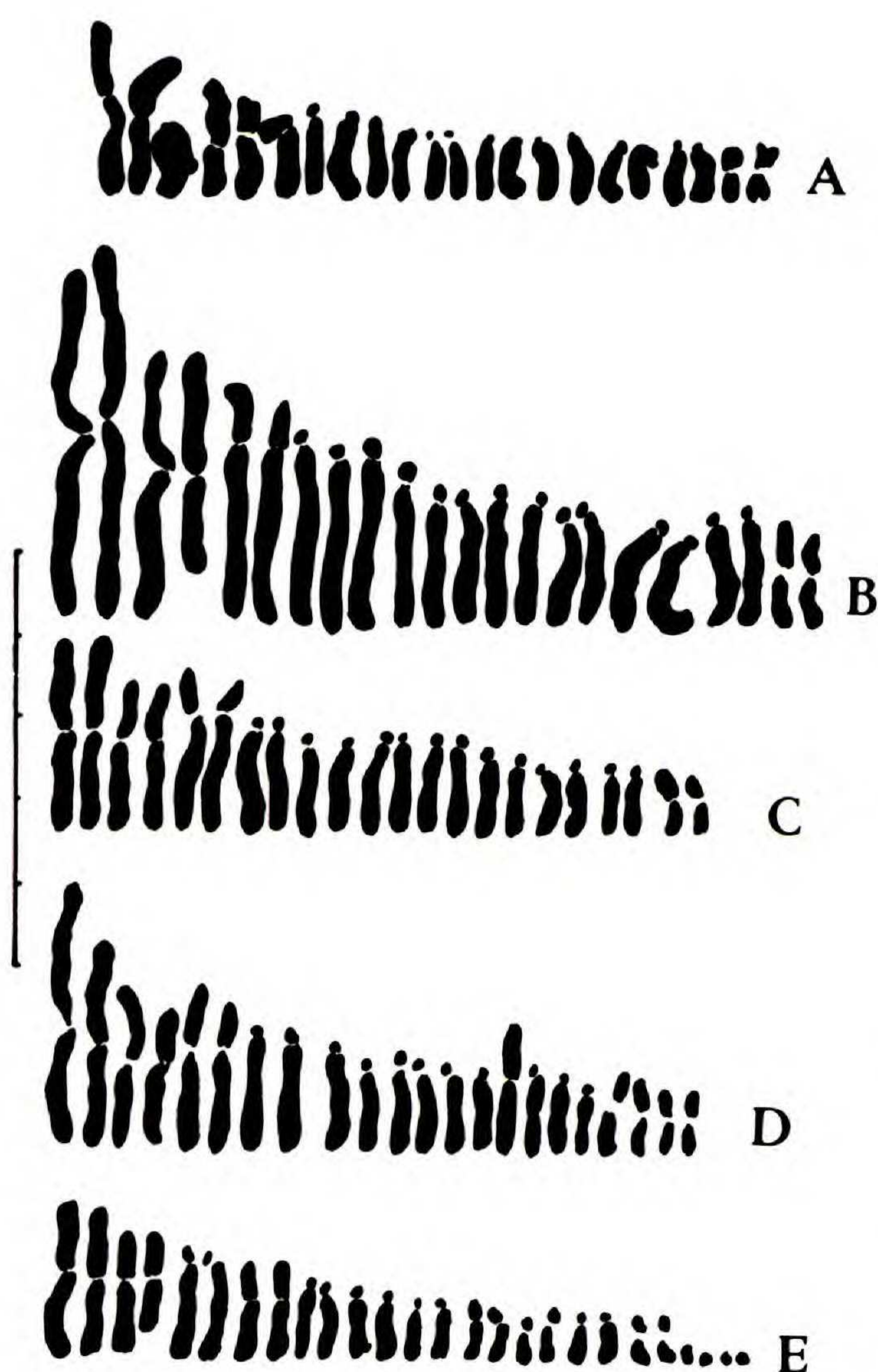


FIGURE 5. Idiograms of the somatic chromosome complements of *Pancratium hirtum* morphotypes.

types. Further deviation is the presence of B-chromosomes in E.

One way of generating genetic variation is by changes in chromosomal morphology, which are reflected in the karyomorphology of the population or the species (Coates, 1979; Coates & James, 1979). Another way is by genic changes that may not be immediately detectable (see Linhart et al., 1981). Thus, while genic changes may not be as immediately detectable as changes in the karyotype, karyotype variation may not be immediately accompanied by morphological divergence.

Karyotype variation in *P. hirtum* is associated with population differentiation and ecological preference. Dickinson & Antonovics (1973) maintained that karyotypic differentiation is a direct response of various biotypes to differing habitat pressures. White (1973) opined that chromosome rearrangements underlie reproductive isolation and,

hence, speciation. It is probable that the ancestral population of *P. hirtum* in West Africa was at some time afflicted by some drastic environmental events that left survivors whose genetic systems suffered some changes. Such survivors occupied different ecological niches to which each had adapted for continued survival. Each has thus become ecologically isolated. Such a situation would be reinforced further by environmental barriers to long-distance pollen dispersal. The smallness of the population that would initially inhabit each ecological niche would enhance both chromosomal evolution and speciation, as asserted by Wright (1940), Bush et al. (1977), and Bengtsson (1980). These events would lead to reproductive isolation and thereby lay the foundation for further divergence of the population, culminating, in time, in the formation of several species. Again, the effects of the environment, especially edaphic factors, may be the major driving force in the karyotypic differentiation in this species. For instance, Morton's (1965) pl. 14, fig. 1 is similar to the karyotypes reported here, but it is not identical with any of them. All of Morton's materials were collected from Ghana. It is therefore possible that other karyotypes may still be encountered within the tropical West African region.

In conclusion, karyotype differentiation in *P. hirtum* has involved: (i) changes in chromosomal morphology resulting from loss or gain of chromosomal segments, which might have been accompanied by changes involving rearrangements of genes and/or gene blocks in inversions and translocations; and (ii) variations in the length of total chromatin material per somatic nucleus. Therefore, it seems likely that ecotype differentiation at the morphological level appears genetically fixed, and this is accompanied by varying degrees of karyotypic change, which presumably originated once the ecotypes had become established.

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